ORIGINAL ARTICLE

NUTRITIONAL VALUE AND GLYCEMIC LOWERING EFFECT OF SPROUTED FERNUGREEK (*Trigonellafoenumgracecum L)* SEEDS

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ABSTRACT

Background: Fenugreek, *TrigonellafoenumgraecumL*, is an annual herb grown in various countries around the world. The seeds are considered important both as food item and medicinal plant. Sprouting is a simple processing method, which involves soaking or steeping dry seeds, cereals or legumes in water until they are saturated followed by germination. **Objective**: The present study investigate the nutritional value and glycemic effect of sprouted fenugreek. **Methodology**: Fenugreek seeds were sprouted for 48 hours. The sprouted and raw fenugreek seeds were analysed for chemical composition, *in vitro* carbohydrate and protein digestibility and antinutritional factors using standard laboratory methods. Thirty male Wister rats were used to evaluate the effect of sprouted and raw fenugreek on weight and their glycemic response. **Results**: The sprouted fenugreek had a higher Protein content (29.33±0.60 %) than the raw fenugreek (26.33±0.65%).The result of the mineral element composition showed significant ($P<0.05$) increase in the levels of calcium, iron, magnesium and zinc in the sprouted fenugreek. Reductions of 71.41% phytic acid and 63.30% tannins were observed in the sprouted fenugreek which led to significant ($P<0.05$) increase in *in vitro* carbohydrate and protein digestibility of the sprouted fenugreek. Decrease in weight of rats was observed both in the group of rats that were fed raw fenugreek and sprouted fenugreek. The weight loss in the group of rats that were fed sprouted fenugreek was significantly ($P<0.05$) more than that of the group that were fed raw fenugreek. The raw and the sprouted fenugreek exhibited glycemic lowering effect but sprouted fenugreek had slightlyhigher glycemic lowering effect. **Conclusions**: The finding concluded that sprouted fenugreek had improved nutritional value and higher glycemic lowering effect.

Keywords: Raw fenugreek, Sprouting fenugreek, Glycemic response

INTRODUCTION

Dietary choice and medicinal herbs remains the basis for maintaining a healthy lifestyle and well-being, especially relating to chronic diseases, which include cancer, cardiovascular diseases (CVDs) and diabetes. Various plants are nutritious and medicinal in nature. Fenugreek is one of such plants that combine these attributes and a well-known spice crop in human diets.[1]

Fenugreek (*Trigonellafoenum-graecum L.*), plant is widely distributed throughout the world and which belongs to the family Fabacecae.[2]The distinctive cuboid, yellow-to-amber colored fenugreek seeds are used both whole and in powdered form. It has been possible to debitter fenugreek seeds by employing various processing methods such as soaking, germination and roasting.[1,3]

The seeds are known to have hypoglycemic and hypocholesterolemic properties. Fenugreek seeds can be a good supplement to cereals because of its high protein, lysine, soluble and insoluble dietary fiber besides being rich in calcium, iron and beta-carotene.[3]

Sprouting, malting and germination are often used interchangeable to describe the process of soaking

or steeping dry seeds, cereals or legumes in water until they are saturated followed by germination under controlled conditions. Sprouting of seeds does not require sophisticated equipment.[4]

The sprouted seeds are outstanding sources of protein, vitamins and minerals and they contain health-maintaining important nutrients like glucosinolates, phenolic and selenium. As the sprouts are consumed at the beginning of the growing phase, their nutrient concentration remains very high.[5]This paper reports on the effect of sprouting on the nutritional value and hypoglycemic response of fenugreek seed

MATERIALS AND METHOD

Source of material

Fenugreek seeds were obtained from Monday Market Maiduguri, Borno State Nigeria. The identity of the plant species was verified by a plant taxonomist at Lake Chad Research Institute Maiduguri, Borno State.

Sprouting of Seeds

One hundred grammes (100g) of fenugreek seeds were cleaned to remove dirt. The seeds were washed three times with water and then soaked (1:3 w/v) for 2 hours at room temperature after which water was drained. The seeds were wrapped in damped cotton cloth. Germination was carried out at room temperature (27°C) over 48 hours. The mouldy seeds were removed by hand and sprouted seeds were washed before sun drying to a constant weight. The dried seeds were ground into a fine powder and sieved using a 1mm pore sieve to obtain a fine powder .[5]

Preparation of raw fenugreek seeds

One hundred grammes (100g) of fenugreek seeds were cleaned to remove dirt, washed and dried to a constant weight. The seeds were ground into a fine powder and sieved using 1 mm pore sieve to obtain a fine powder.[6]

Chemicalcomposition

Proximate analysis was determined by the standard methods.[7] Moisture, ash, protein, fat and fiber contents of the raw and sprouted fenugreek seeds were determined. The carbohydrate content of the samples was calculated using this formula:[8]

% carbohydrate = 100 – (% moisture + % ash + % protein + % fat + % crude fibre).

Energy was calculated by the Atwater conversion factors.[9]

1 g of carbohydrate = 4Kcal.
1 g of fat = 9 Kcal.
1 g of protein = 4 Kcal.

Atomic absorption Spectrophotometer (AAS) AA series (6800 series Shimazo Corp) was used for determination of Ca, Fe, Zn and Mg.

Digestibility

The method of Shekib (2009) was used by employing alpha amylase for *invitro* carbohydrate digestibility.[10]*In vitro* protein digestibility of the samples was determined according to the method described by Nills (1979)by using trypsin.[11] The nitrogen content was determined by micro-Kjedahl method .[7]

Antinutritional Factors

Tannin content of the raw and sprouted fenugreekseeds were determined by the method described by Price and Butler (1977).[12]Phytic acid content of the raw and sprouted fenugreek seeds were determined according to the method described by Davies and Reid (1979).[13]

Weight experiment:

Thirty (30) male albino Wister rats weighing between 100-130gwere purchased from the Animal House, Department of Biochemistry, University of Maiduguri, Borno State, Nigeria. The rats were kept in polypropylene plastic cages and maintained at normal and standard laboratory conditions of temperature (28±2°C) and relative humidity (46±6%) with 12-hour light-dark cycle and adequate ventilation to acclimatize to their environment. The rats were initially fed with commercially available growers mesh and water ad libitum during the period of acclimatization. The rats were grouped into three groups of ten rats each. Group 1(control) was fed growers mesh, Group 2 was fed 100g of raw fenugreek seeds mixed with growers mesh, Group 3 was fed 100g of sprouted fenugreek seeds with growers mesh. The rats were weighed weekly and the experiment lasted for 28 day. This study using experimental animals was conducted in accordance with the international accepted principles for laboratory animals.[14]

Measurement of blood glucose response:
After the 28 days, the animals were fasted for 12 h (overnight fasting). The blood glucose of the animals were taken at zero time from the tail vein before being fed with 2.0 g of the raw fenugreek, sprouted fenugreek and glucose (control), which were consumed within 25 minutes. After the consumption, the serum glucose levels of the animals in each group were taken at 15, 30, 60, 90 and 120 min. intervals using an automated glucose analyser (Accu-chek Active' Diabetes monitoring kit) Roche Diagnostic, Indianapolis, USA.

Data Analysis
The data obtained were analysed using Analysis Of Variance (ANOVA).Duncan's multiple range tests was used to compare differences between the means, using SPSS 11.0 software (SPSS, Chicago, Ill, USA). Significance was accepted at $p \leq 0.05$.

RESULTS
Chemical Composition
Table 1 presents the proximate composition of raw and sprouted fenugreek. Significant (P<0.05) differences were observed in the percentage moisture, protein, carbohydrate and energy values of the raw fenugreek and sprouted fenugreek. The fat, fiber and ash contents did not show any significant (P>0.05) difference. The result of the mineral element composition is presented in table 2.The sprouted fenugreek exhibited significant (P<0.05) increases in the levels of calcium, magnesium, zinc and iron.

Antinutritional Factors
A reduction of 71.41% in the phytic acid content was observed (table 3) with the raw fenugreek having higher values (7.80+0.52 mg/g) than the sprouted fenugreek (2.23+0.20 mg/g). The tannin content also exhibited a decrease of 63.30% with sprouted fenugreek having lower values (2.11+0.33) and the raw fenugreek having higher values (5.75+0.20).

Digestibility
The *in vitro* carbohydrate digestibility of the raw and sprouted fenugreek is illustrated in figure 1. Significant (P<0.05) differences were observed in the raw and sprouted fenugreek. The sprouted fenugreek had higher digestibility than the raw fenugreek. Figure 2 shows the *in vitro* protein digestibility of the raw and sprouted fenugreek. Significant (P<0.05) differences were observed between the raw and sprouted fenugreek. The sprouted fenugreek showed higher digestibility than the raw fenugreek. In both the *in vitro* carbohydrate and protein digestibility, digestibility increased with time.

Effect of Fenugreek on Weight of Rats
A significant (P<0.05) steady increase in weight was observed in the group of rats that were fed the control diet (growers mesh) as shown in figure 3. The group of rats that were fed with raw fenugreek + growers mesh and the group of rats that were fed sprouted fenugreek + growers mesh showed significant(P<0.05) reduction in weight. The group that were fed sprouted fenugreek + growers mesh exhibited lower weight than the raw fenugreek=growers mesh group.

Glycemic lowering effect of fenugreek
The two hour post prandial blood glucose level is presented in figure 4.The initial blood glucose levels (0 mins) are: control group (4.07±0.86mmol/l), raw fenugreek (4.08±0.76mmol/l) and sprouted fenugreek (4.02±0.87mmol/l).The control group (glucose) had higher blood glucose levels (at 15,45,30, 60 and 120 mins) than the raw fenugreek and the sprouted fenugreek groups. The peak blood glucose level of the control group at 45 minutes is 5.30±0.96mmol/l while for raw fenugreek and sprouted fenugreek is 4.81±0.68mmol/l and 4.64± 0.86mmol/l at 45 minutes respectively. The group of rats fed sprouted fenugreek had slightly lower blood glucose levels at 60 mins (4.16 ± 0.65 mmol/l), 90 mins (4.10±0.77mmol/l) and 120 mins(4.01±0.84mmol/l) than the group that were fed the raw fenugreek 4.55±0.61 (60 mins) ,4.19± 0.93 (90 mins) and 4.06± 0.85 (120 mins). Hence the group that was fed sprouted fenugreek exhibited higher glycemic lowering effect than the group that was fed raw fenugreek though the difference was not much.

Table 1: Proximate Composition of Raw and Sprouted Fenugreek

Parameters	Raw Fenugreek	Sprouted Fenugreek
Moisture (%)	6.50±0.30[a]	5.40±0.80[b]
Protein (%)	26.33±0.65[a]	29.33±0.60[b]
Fat (%)	5.00±2.00[a]	4.63±0.73[a]
Fibre(%)	15.00±0.50[a]	14.87±0.051[a]
Ash (%)	3.33±0.66[a]	3.00±0. 40[a]
Carbohydrate(%)	43.84±0.21[a]	42.77±0.87[b]
Energy(Kcal)	325.68±0.60[a]	330.07±0.62[b]

Values are means ± SD of three determination, values in the same row with different superscript are significantly (P<0.05)different

Table 2: Mineral Element Composition of Raw and Sprouted Fenugreek

Parameters (mg/100g)	Raw Fenugreek	Sprouted Fenugreek
Ca	61.90±0.51[a]	66.03±0.18[b]
Mg	19.27±0.22[a]	22.10±0.51[b]
Zn	4.98.09±0.51[a]	7.63±0.33[b]
Fe	5.76±0.19[a]	7.23±0.54[b]

Values are means± SD of three determination, values in the same row with different superscript are significantly (P<0.05) different.

Table 3: Antinutritional Factor Contents of Raw and Sprouted Fenugreek

Antinutrients	Raw	Sprouted
Phytic acid (mg/g)	7.80+0.52[a]	2.23+0.20[b]
Percentage decrease	71.41	
Tannin (mg/g)	5.75+0.20[a]	2.11+0.33[b]
Percentage decrease (%)	63.30	

Values are means ± SD of three determinations, values in the same row with different superscript are significantly (P<0.05) different.

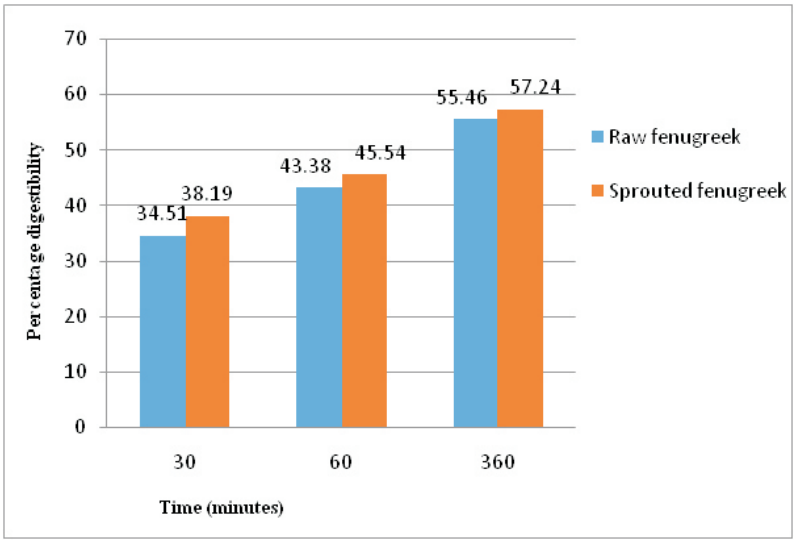

Figure 1: *In vitro* carbohydrate digestibility of raw and sprouted fenugreek

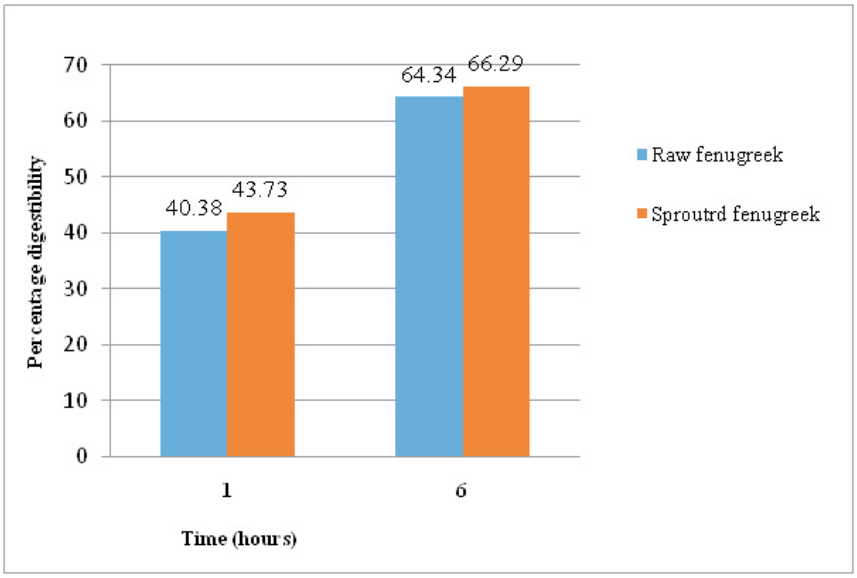

Figure 2: *In vitro* protein digestibility of raw and sprouted fenugreek

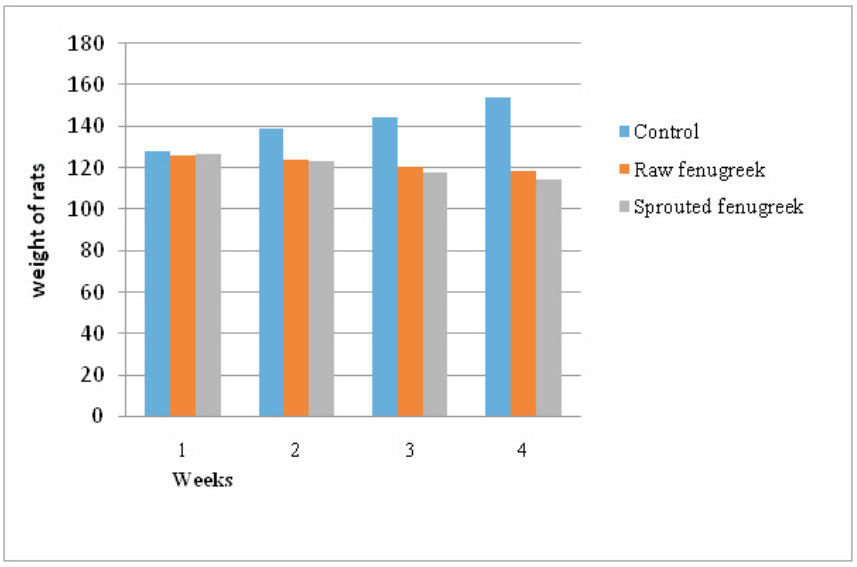

Figure 3: Effect of fenugreek on weight of rats

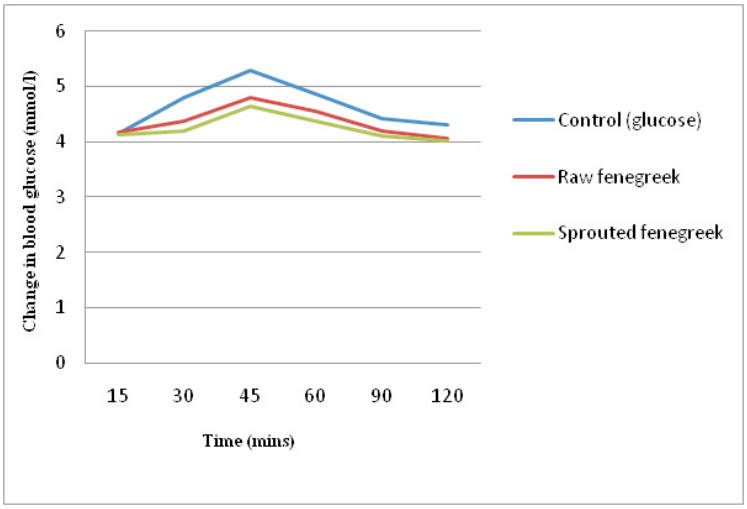

Figure 4: Glycemic lowering effect of Raw Fenugreek and Sprouted fenugreek fed to rats

DISCUSSION

Chemical Composition

Low moisture content is a desirable phenomenon, which increases storage period and reduces microbial activity[15] .Processing techniques such as sprouting improve the nutritional quality of the food products, particularly in terms of protein content.[16]During sprouting the water-soluble protein content increased.[17]Sprouted fenugreek had lower carbohydrate content (42.77±0.87) than the raw fenugreek (43.84±0.21). Raw fenugreek contained15.00±0.50 % fiber, while sprouted fenugreek had a slightly lower fiber content (4.63±0.73).An enzyme α-galactosidase from germinating fenugreek seeds partially attacks galactomannan to yield galactose. The decrease in the polysaccharide and mucilage content may be attributed to their breakdown and utilization by the growing sprouts.[3]The lower carbohydrate content exhibited by the sprouted fenugreek could be due to the utilisation of fat and carbohydrate for biochemical activities of the germinating seeds.[18] γ-Amylase present in the sprout degrades partially the starch thereby leading to a decrease in carbohydrate content. [19]

Significant ($p<0.05$) increases were observed in the calcium, magnesium, zinc and iron content of sprouted fenugreek (table 2). In quantitative terms, this means the process of germination can change a diet of lowiron bioavailability into a diet of intermediate to high iron availability.[20]Increase in the levels of calcium, iron and zinc during germination could be as a result of increased activity of the enzyme phytase. The enzymes hydrolyse the bond between protein-enzymes-mineral to free these essential minerals .[21]

Antinutritional Factors

Phytic acid in plant foods forms complexes with essential dietary minerals such as Ca, Fe, Zn, and Mgand reduces their bioavailability for absorption. Increase in phytase activity on germination led to catabolism of phytic acid. Phytases, or myo-inositol hexaphosphatephosphohydrolases, are enzymes that hydrolyze myo-inositol 1, 2, 3, 4, 5, 6,-hexakis (dihydrogen phosphate) to myo-inositol and inorganic phosphate and thereby increasing the in vitro availability of divalent minerals. [22, 23]

In addition, complex formation of phytic acid with proteins may inhibit the enzymatic digestion of the protein. Raw fenugreek seed flour contained higher amount of phytic acid (7.80+0.52mg/g) and lower amount of tannins (5.75+0.20 mg/g) as shown in table 3. Reductions of 71.41% phytic acid and 63.40 % tannin were recorded.

Digestibility

The increase in *in vitro* carbohydrate digestibility may be due to the degradation of starch into smaller fragments and formation of reducing sugars during germination.[24] Sprouting causes mobilization of protein with the help of protease leading to the formation of peptides, oligopeptides and free amino acid thereby leading to increase in in vitro protein digestibility .[25,26] In addition, the reduction of antinutrients may have contributed to the improvement in protein digestibility. [27]

Effect of Fenugreek on Body Weight

The Loss of weight was observed in the raw and sprouted fenugreek. The loss of weight recorded in the sprouted fenugreek was slightly higher than that of raw fenugreek. Also, fenugreek seeds contain a high proportion (40 %) of soluble fiber. This fiber forms a gelatinous structure which may have effects on slowing the digestion and absorption of food from the intestine and creates a sense of fullness in the abdomen, thus suppresses appetite and promotes weight loss.[28]

Glycemic lowering effect of Raw and Sprouted Fenugreek

The glycemic lowering effect of fenugreek is thought to be largely due to its high content of soluble fiber which acts to decrease the rate of gastric emptying, thereby delaying absorption of glucose from the small intestine.[29]

CONCLUSION

The results obtained shows that the sprouted fenugreek contain appreciable amount of protein, fiber, minerals and higher digestibility. The raw and the sprouted fenugreek exhibited glycemic lowering effect but the sprouted fenugreek had a higher glycemic lowering effect than the raw fenugreek. Hence, sprouted fenugreek may be suitable to be used in fortification of foods for individuals at risk of diabetes.

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